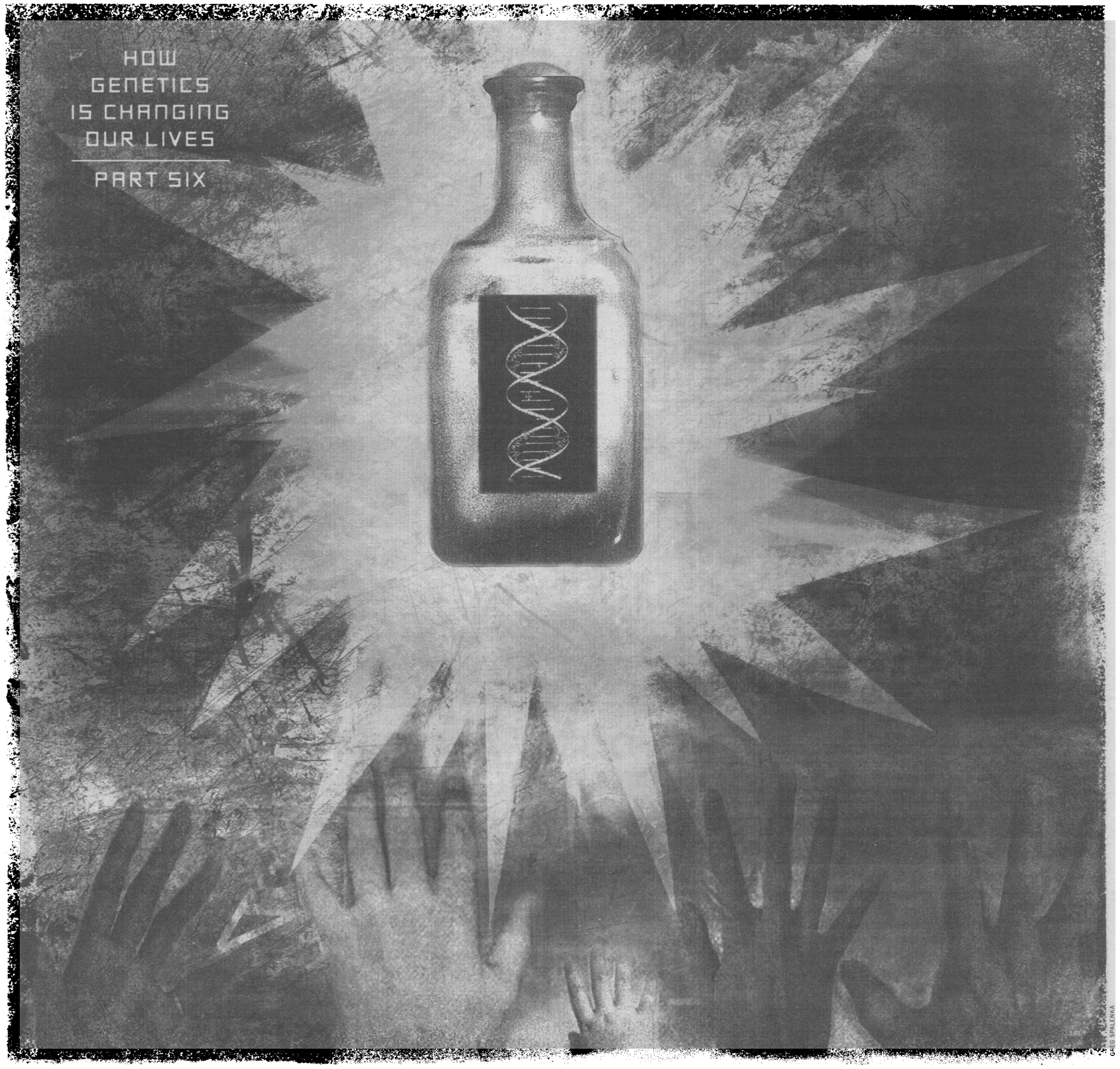
THE DNA DRUGSTORE

Seven years ago, Damian Ivanof's right foot started to hurt. "It's probably arthritis," he remembers a doctor telling him. But as the months passed, "my foot started getting cold and changing colors," and the pain seeped "up toward my knee."

Finally, Ivanof was diagnosed with critical limb ischemia, a mysterious disease that clogs the arteries leading to the legs. It's a gruesome ailment: The lower leg slowly turns black and shriveled, ulcers expose tissue and even tendons, and finally gangrene sets in. During the whole process, the pain is excruciating.

Doctors did a bypass operation, inserting a synthetic artery. That worked—for eight months. A new clot (continued on page 58)



Scientists Are Using Genes to Attack Everything From Cancer to Aging | BY MARK SCHOOFS

SCHOOFS

CONTINUED FROM PAGE 55

forced another operation, then another and another. Ivanof endured eight bypasses, as well as many lesser procedures—angioplasties to clear obstructions; shunts to prop the veins open; IV drips to melt away clots. None of it worked, and his doctor said the next step would be amputation.

That's when Ivanof heard about an experiment being conducted by Jeffrey Isner, chief of vascular medicine at St. Elizabeth's Hospital in Boston. The idea was to inject patients with a gene that makes a protein called vascular endothelial growth factor, or VEGF, which orders new blood vessels to grow. Hopefully, it would induce what one researcher calls a "bio-bypass"—new blood vessels that grow around the clot and revive the dying leg. On January 10, Isner gave Ivanof four shots, one above the knee and three below it.

"The first week I noticed something," Ivanof says, "but I thought it must be in my mind. The second week the pain went down, and I noticed my foot getting warmer. By the third week I didn't have to take any pain medicine. It was the first day in six years I hadn't taken Percocet. By the fourth week, the color in my leg had changed from ashy gray to pink." Suddenly, Ivanof could sleep the night and walk without crutches. "But I still couldn't believe it—to get better in four weeks after six years of pain?" Then Ivanof saw the angiogram, a sort of X-ray for veins. "Before I had one little artery going to the knee, and from there a thin sliver going to the ankle. But now I could see two or three other arteries going down the leg, and little peripherals branching off them."

OF THE 21 PATIENTS who have received this gene therapy, about three-quarters responded, says Isner. Eight have been saved from amputation.

These recoveries prove a simple but astonishing principle: Genes are drugs. Indeed, DNA is the largest and most powerful pharmacy imaginable. Its products—proteins and enzymes—repair wounds, attack infectious invaders, and destroy diseased or cancerous cells every day. Not only that, but sealed in the double helix is the ability to create every cell in the body. In Ivanof's case, scientists figured out how to tap this potential—and that is the future of medicine.

"Gene therapy" is the common phrase for deploying genes as drugs, though a few researchers prefer the more cautious term "gene transfer," which doesn't implicitly promise a benefit. They point out that Isner's experiment is by far the most successful of more than 200 human trials.

"We're years, if not decades, from being able to do true genetic surgery, where you go in and repair a genetic defect," says W. French Anderson, a trailblazer in the field. But he's more bullish than he was in 1958 when, as a senior at Harvard, he first conceived of gene therapy, and more optimistic than in 1990, when he helped conduct one of the first trials in a patient. "Two billion dollars a year is being spent now on gene-therapy research," Anderson points out. "I can't believe we won't have really exciting results over the next five to 10 years."

Already, the investment is paying off. The first large-scale trials of a gene therapy (designed to fight brain cancer) are under way, and just last month the Food and Drug Administration approved a new type of genetically engineered cancer therapy. It's an antibody that stimulates the immune system to attack non-Hodgkin's lymphoma, avoiding the devastating side effects of chemotherapy. Also last month, researchers announced that they successfully treated diabetes in rats by inserting a gene that produces insulin. Introducing this gene would eliminate the need for diabetics to inject insulin before every meal. Just last week, scientists announced they had inserted a gene that corrects for a mutation known to cause obesity, transforming mice from fat to svelte.

The potential is staggering. To fight AIDS, for example, researchers are inserting genes into immune-system cells that would make them capable of neutralizing the virus. But gene therapy's greatest potential probably lies in assailing illnesses that arise within the body itself. Inherited disorders such as sickle-cell anemia and cystic fibrosis could be reversed by correcting the genetic flaws that cause them. And genes can also be directed against ailments—such as heart disease, diabetes, and cancer—that occur because of accumulated genetic damage or aging. In fact, Isner and other teams hope to use VEGF to induce heart bypasses, eliminating the need for invasive surgery. The treatment has already worked in animals.

Ultimately, Ivanof's recovery represents more than just a successful therapy. It's an assault on aging itself, because it proves that "we can regenerate blood vessels" that deteriorate over time, says William Haseltine, CEO of the biotech company Human Genome Sciences Inc., which is collaborating with Isner. Such "regenerative therapy" will become more common, Haseltine says, and by using it on more and more organs, scientists could "extend the aging process very dramatically."

OF THE 222 GENE-THERAPY trials in the United States, about three-quarters focus on cancer. That makes biological sense—cancer occurs because the genetic program goes haywire—and it also makes economic sense. Although there are about 5000 inherited genetic diseases, such as Huntington's chorea and hemophilia, all of them together constitute only 5 per cent of America's total disease caseload. "So," says veteran gene-therapy researcher Nelson Wivel, "this is not something to light up the heart of a drug company." But there's money in cancer.

Whether the ultimate cause is cigarette smoke or random internal damage, cancer is largely a disease of aging: It emerges when a cell accumulates so many "genetic lesions"—a medical term for harmful mutations—that the cell escapes the body's intricate control mechanisms and grows wildly. Normally, most cells die after they have divided 60 to 100 times. But cancer cells are immortal, capable of dividing and growing forever. In 1951, Henrietta Lacks died of cervical cancer. But a researcher harvested cells from her tumor. Dubbed HeLa cells, they are now in petri dishes all over the world, dividing as vigorously as ever.

As a tumor grows, its cells keep mutating, diverging from one another. Some ovarian tumors, for example, contain hair and teeth. This diversity makes cancer a formidable enemy, because one treatment might kill off only some of the cells, leaving others to proliferate. And cancer cells, like viruses or bacteria, mutate to become drug resistant. Sometimes cancer cells evolve so that they actually expel chemotherapy drugs. "Cancer is so horribly complicated," says Wivel, "and all we're doing with gene transfer is manipulating little tiny facets of it."

Still, there's more hope than ever before, because researchers have identified about 100 genes that seem crucial to cancer. When oncogenes, such as *ras*, get turned on too strongly, cells start growing out of control. When that happens, tumor-suppressor genes, such as *p53*, are supposed to trigger a last-ditch mechanism called programmed cell death—but in many cancers, *p53* is disabled.

Jack Roth, a doctor at Houston's M. D. Anderson Cancer Center, is trying to put functional *p53* back into cancer cells. Researchers took the adenovirus, which causes the common cold, stripped it of its ability to cause disease, and loaded it with the *p53* gene.

That's different from Ivanof's case. He was injected with "naked DNA," in which the gene was unaided by any delivery system. That approach often fails, because genes must get inside a cell's nucleus to function, and cells often won't take in foreign DNA. So scientists usually

CONTINUED ON PAGE 61

SCHOOFS

CONTINUED FROM PAGE 58

enlist viruses to do what they do best: invade the nucleus, but now armed with a therapeutic gene. Some researchers are even experimenting with HIV, engineering it to be a harmless carrier of therapeutic genes.

The Holy Grail is a delivery vehicle that could be injected in the arm like a vaccine and that would seek out and enter only specific cells—all without provoking the immune system to attack it. But science is nowhere near that: Roth's team injected their *p53*-carrying virus directly into the tumors.

The 86 patients—with head, neck, and lung cancer—were late-stage. Some of the tumors, says Roth, "were as big as a large orange." Even so, "one-third showed clinical benefit," and a few have "been in remission for the past six months." Larger trials have just begun.

This isn't as dramatic as sprouting new blood vessels and avoiding amputation. But cancer is a war of attrition, says leading gene-therapy researcher R. Michael Blaese: "If you have a cancer therapy that 25 per cent of patients respond to, and you add a new treatment that 35 per cent respond to, that's a big deal." Adding *p53* restores just one cancer-fighting mechanism—and some tumors might mutate in ways that thwart it. Doctors of the future may analyze a tumor's damaged DNA and then administer a "cocktail" of therapeutic genes.

CANCER IS ONE OF five age-related diseases that account for the vast majority of deaths between ages 50 and 85. The others are diabetes, Alzheimer's, stroke, and heart disease. "All of these have a strong genetic component," says Rudolph Tanzi, an expert on aging who helped discover the three known genes for early-onset Alzheimer's.

In some cases, inherited gene defects lead to disease virtually 100 per cent of the time. But in most, genetic differences merely make people more susceptible. For example, 30 per cent of Americans are born with a particular mutation that seems to predispose them to Alzheimer's.

How could deleterious genes be so common? The answer lies in a statistic: At the beginning of this century, the average life span in America was 47. Now it's 76. "Genetic variations that used to be neutral or innocuous suddenly make a difference when we start living past 60," says Tanzi. Natural selection couldn't have eliminated these variants, because "humans didn't live long enough to get these diseases."

This leads to what Tanzi calls "a weird paradox. For the first time, we've been able to extend our life span so much that we have outpaced our own genome." Simply put, we weren't designed to live this long, and the big five age-related diseases are the proof of that. But, says Tanzi, "we don't have to be slaves to our DNA."

Of course, once the average life span reaches 100 or more, new genetic diseases that would never have had a chance to develop will suddenly spring up. "We'll be playing this cat-and-mouse game endlessly," says Tanzi. Still, he believes that "by the middle of the next century, we will expand our life span by a decade for every decade of research."

"Extremely far-fetched" is what Richard Hodes, director of the National Institute on Aging, calls this estimate. But even he concedes, "It's presumptuous to say that, genetically, it can't happen."

Haseltine goes further. "Some organisms, such as bacteria, are immortal. They grow by dividing, so who's to say what's parent and what's daughter?" From this perspective, DNA itself is 4 billion years old, and every living organism is just a husk, discarded as soon as DNA no longer needs it. But, insists Haseltine, "there is nothing intrinsic about living matter that says it has to die." Rather, life span is just another evolutionary adaptation.

Natural selection "builds an organism to last until it would otherwise starve, get eaten, or be squashed. If a mouse were built to live as long as a human, it wouldn't. It would get killed. So there's no point in making a mouse that could live to 80." Much better, he says, to take the energy needed to repair aging cells and invest it in food gathering or reproduction. "Age," Haseltine concludes, "is determined by our ecological niche." But now that humans are in a position to steer evolution, "the goal is to attach our conciousness to DNA for a longer ride."

For starters, he says, "we can learn to replace our tissue." Bone marrow contains stem cells, which produce every type of blood cell. "There's no reason you can't implant stem cells and regenerate a whole new blood system. It can be done, it is being done. In cancer, sometimes a 50-year-old gets bone marrow from a 20-year-old donor." Those patients have hybrid ages.

In the future, organ transplants will give way to organ replacement, says Caleb Finch, an expert on aging at the University of Southern California. Doctors will "take a skin cell from a person, put it in culture, and grow up a kidney," he says. "That's not pie in the sky. This thought is in the minds of thousands of scientists."

The biotech company Geron is betting on a gene that apparently helps cells become immortal. Chromosomes come with protective caps at their ends, called telomeres. They've been compared to the plastic tips on shoelaces, protecting the chromosomes from wear and tear. But each time a cell divides, the telomeres get a little shorter. When they are completely worn down, cell division begins to damage the chromosome and its precious genetic material. At this point, the cell stops dividing and heads toward death.

But certain cells, such as stem cells, are spared this fate. Their telomeres are continually repaired by a special enzyme called telomerase. Last summer, scientists isolated and cloned the gene for telomerase, and just this month they succeeded in turning it on in cells. Now, four research teams are racing to answer the ultimate question: Will telomerase give cells a second youth?

The answer should come by the end of January—and if telomerase works, one of Geron's first targets will be the immune system, which is often devastated by cancer treatments. Doctors might remove immune-system cells, genetically engineer them to express telomerase, and then put them back in the body. Alternatively, researchers might find the genetic switch that keeps telomerase off and find a drug to inhibit this "repressor mechanism." Or they might load the telomerase gene onto a virus that will carry it into cells.

"We're not trying to make an immortal human," says Geron's chief scientist, Calvin Harley, just extend people's "health span." Indeed, no single gene controls aging. The process is at least as complex as cancer, and nongenetic factors play a huge role. The bristlecone pine can reach 5000 years on the highest peaks but rarely more than 1000 years farther down the mountain, and this century's dramatic rise in human life span was achieved not by any genetic change but by improving sanitation, introducing antibiotics and vaccines, and enhancing public health.

But one role genes play is to repair damage caused by the environment. Just last month a gene was discovered in mice that apparently defers a host of age-related diseases such as arteriosclerosis. Researchers named it *klotho*, after the Greek goddess who spins the thread of life.

As scientists unravel the secrets of how DNA protects itself and the cells that carry it, says Finch, they will devise "real interventions at the molecular level of aging." And that, he declares, will be "political dynamite."

"Suppose a small percentage of the population, because of connections and wealth, were living to 120 or 140, while most of those around them were still in the trenches, subject to the old life span. It could create whole new health-related castes. How generally these medical marvels will

CONTINUED ON PAGE 64

SCHOOFS

CONTINUED FROM PAGE 61

be available and what they will cost—that's a political question the industrial democracies are going to be confronted with pretty soon."

FOR NOW, THE PROBLEMS are more personal than political, because genetic medicine is in its infancy. In fact, it can be downright crude.

William Coughlin has a particularly vicious brain cancer called glioblastoma. Even with surgery, radiation, and chemotherapy, life expectancy is one year. Coughlin is 39 and has a son who's not quite two years old. "My father died when I was nine," says Coughlin, "and looking at my son, well, I don't want him to lose his father." So Coughlin seized "my only hope."

This September, doctors surgically removed the tumor from his brain and genetically altered its cells. Then they put these tumor cells into "diffusion chambers," medical tea bags that keep the cancerous cells in but allow the small molecules they secrete to diffuse out. Doctors cut open Coughlin's stomach and let the tumors steep inside him for 24 hours.

The procedure seems almost reminiscent of medieval bloodletting. But even more shocking is that researchers don't know what molecule the altered cancer cells discharge. All they know is that their procedure—which they won't discuss because a patent is pending—makes the cells secrete something that apparently causes tumors to shrink. "We think we have identified a trigger, but we haven't identified the gun yet," explains clinical investigator David Andrews, of Philadelphia's Thomas Jefferson University Hospital.

For the moment, Andrews is very excited about his tea-bag trial. Four patients before Coughlin responded in ways that he says are "truly remarkable." But many of these "remarkable" responses were seen in autopsies—the other four patients are dead. Their tumors grew back, says Andrews, because they were "end-stage patients."

Coughlin is not so far along, and for the moment, his cancer has receded. Coughlin saw the MRI scans of his tumor, before and after treatment. "It looked like a collapsing supernova," he says, adding that the tumor has shrunk by about one-third. So will he, like Ivanof, be rescued by genetic medicine? Probably not, says Andrews: "I'd have to say he's still terminally ill." He compares the procedure to the first flight at Kitty Hawk, saying, "This is not a cure. But if there were a strategy to lead to a cure, this is it."

Doctors have the luxury of getting excited over small signs of progress. But what about patients? If the therapy doesn't work—if the tumors start growing again and Coughlin's son is left fatherless—would Coughlin feel betrayed? "No," he replies. "If, God forbid, I go, I'll have given them more knowledge. And they'll perfect it." V

Part Seven: The Future

Research assistance: Mina Seetharaman and Catherine Donaldson-Evans

THE DRUG PHARM

"THE MAGICAL PROPERTY of genes is that they're seeds," explains William Haseltine, who runs the company Human Genome Sciences. "Cultivate them and they grow," producing proteins and enzymes, the chemicals that carry out every function in the body. With gene therapy, doctors transfer a gene into a patient, where it then "grows" the protein. But often, the protein itself can be given as a drug.

For example, Haseltine's company has found a protein that heals wounded skin. Pictures tell how effective it is: Skin, dosed with steroids so it couldn't heal, was cut. Ten days later, the incision that didn't get the healing protein still gapes open. But the treated wound—given just one topical application of the protein—is completely mended with new, pink skin.

Protein molecules are far too large and complex for labs to synthesize, and only minute amounts can be harvested from humans, so they used to be unavailable. But now, the gene that makes them can be cloned and inserted into plants, animals, or even insects, which will then produce large quantities of the protein. Several such bio-drugs are being "pharmed," and many more are in the pipeline.

Turning genes or their proteins into drugs is probably the ultimate medical use of DNA. But genetics also reveals the chain of events that leads to disease. Consider the new AIDS wonder drugs called protease inhibitors. Back when HIV was discovered, one of the first things Haseltine and other scientists did was sequence its genes. Then they deleted each gene to see what it did and whether HIV could survive without it. One gene made an enzyme called protease, which HIV needs to replicate. Pharmaceutical companies started testing compounds to find some that would disable the enzyme—and a decade later, protease inhibitors are giving AIDS patients real hope.

This same strategy can be used to fight ailments that arise from the body's own DNA. But HIV has only 10 genes, whereas the human body has an estimated 100,000. Which are the culprits? To find out, Haseltine's researchers compare genes used in normal and diseased cells.

For osteoporosis, they found a gene that is used *only* in the destructive cells, the ones that degrade bones. Then they checked their computers and discovered that the gene is closely related to a mouse gene that makes a protein known to degrade cartilage. Suddenly, they had a target for drug therapy: Block this protein and doctors might be able to halt the disease that bends old ladies into pretzels.

"Now is like AIDS in 1988—the basic discoveries have been made, but the drugs are seven or eight years away," says Haseltine. "We've transferred 8000 genes to our partners"—mostly large pharmaceutical companies such as SmithKline Beecham—"and initiated hundreds of drug discovery projects."

At the heart of Haseltine's company is an L-shaped assembly line. Down one corridor, machines sequence DNA and pour the data into computers, 24 hours a day. Down the other corridor, robots perform 10,000 biological assays daily, testing genes to find out exactly what they do. The result: When the company's scientists want to know about a particular gene—where in the body it functions, and how—they point and click. "The computer screen," says Haseltine, "is our microscope."

As a CEO, Haeltine is as famous for salesmanship as for science. Though few firms can boast as rich a genetic database, others do use a similar approach. And as the government's Human Genome Project puts our entire genetic code on the Internet, more and more information about how those genes function will enter the public domain.

This project has already made biology easier and faster. In a recent interview with *Bioworld Today*, Nobel laureate Thomas Cech explained how it took "three years to purify, sequence, and clone the gene" for an enzyme in a protozoan. "And then it took a few minutes to leapfrog . . . to the human [gene], using the computer and the Human Genome Project." —M.S.